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Research article

A rapid and economical method for the maceration of wood fibers in *Boswellia serrata* Roxb.

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Abstract: *Boswellia serrata* Roxb. (Burseraceae) is endemic to India and naturally distributed to Chhattisgarh, Madhya Pradesh, Maharashtra, Odisha and parts of Rajasthan. It is drought tolerant and resists fire. It has high pharmaceutical value due to its gum-resin. A rapid, convenient and economical method for complete maceration of its wood fibers was developed during the process of fiber analysis. Wood core samples were collected from the trees of *B. serrata* growing in Institutes' campus and their fibers were macerated using different concentrations of macerating agent (nitric acid). 50% nitric acid was found very efficient to separate out all the fibers. This maceration protocol resolves clearly all the compact fibers and made their measurement convenient. The method may be useful for studies of wood fibers for exploration of better populations and may be used for quality paper production.

Keywords: Boswellia serrata - Nitric acid - Wood fiber - Maceration.

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INTRODUCTION

Since ancient times, wood of many trees is regularly in use for quality furniture and in pulp and paper industry. Durability and strength of products depends upon tensile strength of wood fibers. Cellulosic fibers are normally longer in sapwood than heartwood and contribute 40–45% wood's dry weight of fiber wall which makes it suitable for pulping. Sapwood of *Boswellia serrata* is deficient in lignin, hemicelluloses and other extractives and thus widely used in paper pulp process. Pulp formation involves mechanical and chemical processing. In mechanical process wood is chopped whereas in chemical process fibers are capable of blending easily without breaking. Besides other properties, the physical dimensions of the fiber are among the most important factors in pulping and other commercial purpose. Fiber length is one of the quality parameters for pulpwood, and it has been extensively studied in relation to tree age and within-tree position (Hudson *et al.* 1995). With increasing interest in non-wood pulping, it is essential to know the fiber length for interpretation of variability also (Han *et al.* 1999).

Anatomical studies of plant stems or other parts, rarely convey an accurate picture of the real nature of the cells of which they are composed. One method which reveals cells in their cellular structure is the dissociation method. The target plant is treated with chemicals which dissolve the middle lamella and allow the cells/fibers to become separated from one another. However, in some plants, the mild maceration process will not completely dissociate to a single fiber unit, resulting in an aggregate of fibers. These aggregates of fibers have the appearance of a single fiber. The maceration process is actually small-scale pulping and sometimes referred "test-tube pulping". Some processes result in maceration as well as bleaching.

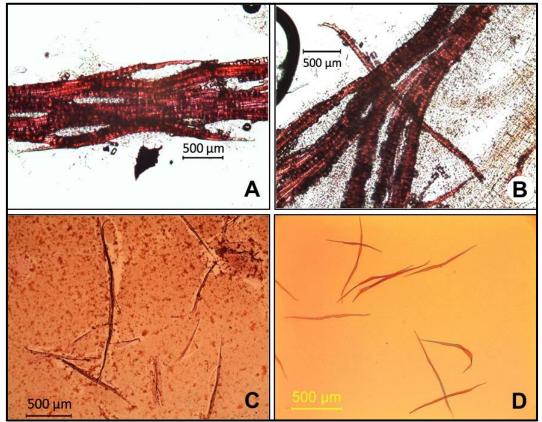
Boswellia serrata (Burseraceae) is an excellent source of pulp for production of very high quality paper. Species is endemic to India and naturally distributed in central Indian deciduous forest. Species has been heavily exploited in past and therefore require conservation and field plantations of its better genotypes in terms of its fibers to overcome its demand in pulp and paper industry. In order to make an analysis of wood fibers of *B. serrata*, an economical, simple and rapid method for maceration of wood fibers was developed which may be

useful for other species also. Fiber maceration technique described by Jorge *et al.* (2000) was also compared in this study.

MATERIALS AND METHODS

The wood core samples were collected by increment boring from the trees of about 20 years old *B. serrata* stand at Institute's campus. Collected wood core samples were submerged in 37% formaldehyde solution. Prior to start the maceration process, samples were drained for formaldehyde solution to avoid more evaporation of fumes. Three concentrations of nitric acid *viz.* 40%, 50% and 60% were used. Wood core samples were taken in test tubes, dipped them completely in nitric acid solution and kept in a water bath at 70°C. Maceration process completes in 5–6 hrs with separation of white colored fibers. Test tubes containing macerated fibers were removed from water bath and allowed to cool at room temperature. After cooling, nitric acid was drained and macerated fibers were washed thrice with distilled water and filtered using Whatman Grade 1 filter paper for separation of fibers. Precautions for well drain of formaldehyde solution is necessary prior to immersion in nitric acid (when formaldehyde preserved samples immersed in nitric acid, the exothermic reaction occurs due to oxidation of formaldehyde).

For slide preparation fibers were stained with 20% safranin solution and again washed with distilled water for destaining of excess safranin. Placed some amount of fiber suspension on a standard glass slide with the help of ink/medicine dropper and allowed for air drying. Mounting was done in canada balsam using a cover glass. Use of glycerol enhances visibility of fibers.



RESULTS AND DISCUSSION

Figure 1. Macerated fibers with: A, 1:1 glacial acetic acid: hydrogen peroxide solution; B, 40% nitric acid; C, 60% nitric acid; D, 50 % nitric acid.

In present study, partial maceration was observed in 40% nitric acid (Fig. 1B) whereas in 50% and 60% nitric acid complete maceration occurs. However, observations recorded under microscope (5X magnification using Leica microsystem EC 3, Switzerland with software LAS 4.3.0) reveal splitting of fibers in 60% nitric acid (Fig. 1C) whereas, 50% nitric acid exhibited separation of fibers without their splitting (Fig. 1D).

Maceration of *B. serrata* samples by the method of Jorge *et al.* (2000) reveals no separation of fibers even after 24hrs dipping in prescribed solution (glacial acetic acid : hydrogen peroxide in 1:1). Only partial maceration observed in samples incubated at 70°C for 3 hrs. Therefore, this method was not found efficient to macerate *B. serrata* fibers and comparatively time consuming and costly also.

Nitric acid acts as an easy and fast resolving agent to break down the middle lamella for separating the cells. Boiled nitric acid separates organs/cells much faster. Results reveal that 50% nitric acid is not only convenient for maceration in hot condition but dissolves other extractives also. This protocol helps to reduce the time and chemicals' cost. Nitric acid in combination with other chemicals has been used for maceration of fibers in different species. Schultze's method describes the application of a combination of various concentrations of nitric acid with a small quantity of potassium chlorate and allowed to stand at room temperature or heated slightly to initiate the reaction (Chamberlain 1915). There are some variations of Schultze's method. Jeffrey (1917) proposed maceration by using mixture of equal portions of freshly combined 8 to 10% nitric acid and chromic acid. Slightly heating hastens the reaction and macerates wood samples. Schmid (1982) made sonification and other improvements on Jeffrey's method. Large (up to pencil-size) pieces of wood are macerated in Jeffrey's solution. Franklin (1945) used acetic acid and hydrogen peroxide. Spearin and Isenberg (1947) used sodium chlorite and acetic acid for maceration and found considerable damage in fiber length. Burkart (1966) described treatment with triethylene glycol containing an organic catalyst, such as phenol sulfonic acid or p-toluene sulfonic acid at 130°C. Hall et al. (1986) used nitric acid and formaldehyde for maceration of fibers. Han et al. (1999) compared fiber length measurement techniques such as digitizing, the Kajaani procedure and NIH image in Kenaf (Hibiscus cannabinus L.) and developed a relationship between fiber length, growth and pulping condition. Arau'jo et al. (2002) performed microwave assisted acid digestion $(2.0, 3.0, 5.0, 7.0 \text{ and } 14.0 \text{ mol}^{-1} \text{ with } H_2O_2 (30\% \text{ v/v})$ for evaluation of residual carbon content (RCC) using inductivity coupled plasma optical emission spectrometry (ICP-OES) with axial viewing.

CONCLUSION

It reveals through our study that the maceration of wood fibers in 50% nitric acid consuming less time and also economical than the other methods. Procedure is not only rapid and economical but also yielded complete maceration which is a prime requirement for wood fiber analysis. Since, the pulp of *B. serrata* provide good strength and quality in paper industry when mixed with 25–40% long fibred bamboo pulp, the procedure may be employed for evaluation of better genotypes and their conservation and multiplication for afforestation programmes to meet out the demand of pulp-paper industry and pharmaceutical industry for better economic returns.

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